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FATTY ACID CONTENT AND COMPOSITION IN RELATION TO GRAIN SIZE OF BARLEY

WALTER DE MAN and PETER BRUYNEEL

Laboratorium voor Toegepaste Organische Scheikunde, Katholieke Universiteit Leuven, Kardinaal Mercierlaan, 92, B-3030, Heverlee, Belgium

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Key Word Index—*Hordeum vulgare*; Gramineae; barley; fatty acids; grain size; genotype.

Abstract—The relationship between grain size and total fatty acid (TFA) content and composition, as well as the contribution of corn size distribution to the variation of fatty acid contents between barley types, were studied in nine barley varieties (six-rowed and two-rowed winter barleys, and two-rowed spring barleys) fractionated according to grain size. Smaller grains contained as much TFA as the bigger ones. On average, the proportions of saturated fatty acids (16:0 and 18:0) and of oleic acid (18:1) are higher in large grains than in small kernels, whereas the percentages of linoleic (18:2) and linolenic (18:3) acids are lower. The higher 18:1 and the lower 18:3 proportions in two-rowed barleys compared to their six-rowed counterparts, can be explained by the indirect effect of the different grain size distribution between the barley types.

INTRODUCTION

Many efforts have been made to establish genotypical and environmental variability in total fatty acid (TFA) content and composition of barley grains [1–6]. The comparisons of barley samples, however, have not included the possible effects of differences in grain size distribution and of a relationship between grain size and fatty acid content and/or composition. Our working hypothesis was that if a relation exists between grain size and fatty acid content, at least part of the observed variability among barley types could possibly be explained by an indirect effect due to a different grain size distribution.

In rapeseed (*Brassica napus*), a higher total lipid content was found in smaller or larger seeds, as well as differences in fatty acid composition between seed sizes [7].

Although in malting and brewing it is known that barley grains of different sizes (widths) differ in nitrogen content and in the rates of water uptake during steeping, germination and colouring during kilning [8], no information is available on the relationship between grain size and lipid composition in barley.

RESULTS AND DISCUSSION

The results of triplicate fatty acid analysis on whole kernels of nine varieties, fractionated according to grain width, are shown in Table 1. The range of 2.9–3.7% found for TFA content of grains of different barley varieties and different grain sizes, does not exceed the ranges reported previously [3–6, 9]. Analysis of variance was carried out on the total dataset and on the data of different barley types separately. The class variables used in the ANOVA model were variety, grain size and the interaction between these factors. Furthermore, for each set the differences between means were evaluated using Duncan analysis (Table 2). Finally, simple correlation coefficients were calculated between thousand corn weight (TCW) and fatty acid composition. Separate statistical analysis of spring barleys shows that TFA content and composition are not influenced much by grain size; there is only significance for the increase of stearic (18:0) and oleic (18:1) acid proportions with greater grain size (Table 2).

On the contrary, grain size is significant in explaining the variability in fatty acid composition of winter barleys, the TFA content remaining constant. Grains with bigger corn size have significantly higher 16:0 and 18:1 proportions and lower proportions of unsaturated fatty acids (18:2 and 18:3) than the smaller grains.

Two-rowed winter barleys have a significantly lower TFA content in the 2.2 mm grain size fraction. The proportions of 16:0 and 18:0 are not significantly affected by grain size, while a positive relationship between grain size and 18:1 proportion and negative relationships with those of 18:2 and 18:3 are found. In six-rowed winter barleys there is no difference in TFA content of grains with different corn sizes. Grains with a bigger size have significantly higher proportions of saturated fatty acids (14:0, 16:0 and 18:0) and lower proportions of unsaturated fatty acids (18:2 and 18:3).

In order to explain the relationships between grain size and fatty acid composition, two possible hypotheses will be discussed below. Bearing in mind the different fatty acid content and composition of embryonal axis, endosperm and hull [10] and the difference in weight proportions of these tissues in grains of different grain width, we tested the hypothesis that variations of fatty acid composition with greater grain size can be explained by the higher weight proportion of endosperm tissue. Simple calculations, using the mean fatty acid composition for each tissue [10], show that this argument is only valid for the explanation of the variation of 18:3 content with grain size. Alternatively, total lipids can be divided into neutral, glyco- and phospholipid fractions, each having different fatty acid compositions. Changing the proportions of these lipid fractions, without altering

Table 1. Grain size distribution (% wt), thousand corn wt (TCW g dry wt), mean total fatty acid content (TFA mg/g dry wt) and fatty acid composition (% TFA) of nine barley varieties, grown at one location in Belgium and fractionated according to grain width (results of triplicate analysis)

					Fatty acids					
Variety	Type	Fraction	% wt	TCW	14:0	16:0	18:0	18:1	18:2	18:3
Sonja	Two-rowed winter	2.2	4.6	28.3	0.2	22.4	0.9	11.4	57.4	7.8
		2.5	12.9	37.4	0.2	23.4	1.0	12.0	55.8	7.5
		2.8	80.3	52.0	0.2	23.1	0.7	13.7	55.3	6.9
Marinka	Two-rowed winter	2.2	4.4	27.2	0.2	22.0	0.8	12.0	57.5	7.6
		2.5	14.1	36.2	0.2	22.7	0.8	12.6	55.9	7.8
		2.8	79.5	50.3	0.1	24.3	0.9	13.1	54.7	6.9
Isabella	Two-rowed winter	2.2	5.8	27.4	0.3	24.3	0.8	10.8	55.9	7.9
		2.5	18.4	35.5	0.2	23.7	0.9	11.4	56.1	7.7
		2.8	73.1	48.6	0.2	28.3	0.8	11.3	52.7	6.7
Gerbel	Six-rowed winter	2.2	9.6	27.4	0.5	27.4	0.9	11.1	52.5	7.6
		2.5	31.0	36.0	0.3	25.2	1.1	11.9	53.7	7.8
		2.8	55.9	47.4	0.3	28.9	0.9	11.8	51.5	6.6
W1261	Six-rowed winter	2.2	17.3	30.2	0.2	23.3	0.7	12.2	55.2	8.4
		2.5	35.5	37.4	0.3	25.5	0.8	12.3	53.5	7.6
		2.8	41.1	48.1	0.6	33.8	0.9	11.4	47.1	6.0
Corona	Six-rowed winter	2.2	9.1	20.7	0.2	22.3	1.0	11.5	57.3	7.7
		2.5	32.4	36.1	0.2	21.3	1.2	12.2	57.0	8.1
		2.8	54.9	47.0	0.2	23.0	1.1	12.0	56.4	7.2
Aramir	Two-rowed spring	2.2	3.3	26.4	0.3	23.5	0.7	12.4	56.8	6.3
		2.5	14.0	32.8	0.3	23.3	0.7	12.3	57.0	6.5
		2.8	81.0	42.6	0.2	21.3	0.9	13.0	57.8	6.8
Triumph	Two-rowed spring	2.2	4.1	26.7	0.2	24.3	0.7	12.2	56.5	6.2
		2.5	20.3	32.7	0.2	26.2	0.7	11.9	55.1	5.8
		2.8	73.7	41.6	0.2	25.0	0.7	12.3	55.6	6.2
Galant	Two-rowed spring	2.2	4.1	25.9	0.2	22.8	0.6	11.6	57.9	6.9
		2.5	17.8	33.4	0.3	24.1	0.7	12.0	56.3	6.5
		2.8	75.3	42.9	0.2	23.3	0.8	12.1	57.2	6.4

Table 2. Mean total fatty acid content (TFA mg/g dry wt) and mean fatty acid composition (% wt TFA) of different grain size fractions

			Fatty acids (%)					
	Grain size	TFA	14:0	16:0	18:0	18:1	18:2	18:3
Total dataset	2.2	33.5 A	0.2 A	23.6 A	0.8 A	11.7 A	56.3 A	7.4 A
(n = 81)	2.5	33.7 A	0.2 A	23.9 A	0.9 B	12.1 B	55.6 A	7.3 A
	2.8	33.4 A	0.3 A	25.7 B	0.9 B	12.3 C	54.3 B	6.6 B
Winter barleys	2.2	33.8 A	0.2 A	23.6 A	0.9 A	11.5 A	55.9 A	7.8 A
(n = 54)	2.5	34.4 A	0.2 A	23.6 A	1.0 B	12.1 B	55.3 A	7.8 A
	2.8	33.7 A	0.3 A	26.9 B	0.9 AB	12.2 B	53.0 B	6.7 B
Six-rowed winter barleys	2.2	32.8 A	0.3 A	24.3 A	0.9 A	11.6 A	55.0 A	7.9 A
(n = 27)	2.5	32.7 A	0.3 A	24.0 A	1.0 B	12.1 B	54.7 A	7.9 A
	2.8	31.8 A	0.4 B	28.6 B	1.0 B	11.8 A	51.7 B	6.6 B
Two-rowed winter barleys	2.2	34.7 A	0.2 A	22.9 A	0.8 A	11.4 A	56.9 A	7.8 A
(n = 27)	2.5	36.1 B	0.3 A	23.3 A	0.9 A	12.0 B	55.9 A	7.7 A
	2.8	35.7 B	0.2 A	25.2 A	0.8 A	12.7 C	54.2 B	6.9 B
Spring barleys	2.2	32.9 A	0.2 A	23.5 A	0.7 A	12.0 A	57.1 A	6.5 A
(n = 27)	2.5	32.3 A	0.3 A	24.5 A	0.7 B	12.1 A	56.0 A	6.3 A
	2.8	32.9 A	0.2 A	23.2 A	0.8 C	12.4 B	56.9 A	6.5 A
Two-rowed barleys	2.2	33.8 A	0.2 A	23.2 A	0.8 A	11.7 A	57.0 A	7.1 A
(n = 54)	2.5	34.2 A	0.2 A	23.9 A	0.8 A	12.1 B	56.0 AB	7.0 A
	2.8	34.3 A	0.2 A	24.2 A	0.8 A	12.6 C	55.5 B	6.7 B

There was no significant difference (Duncan) between the means in the columns coded by the same letter ($\alpha < 0.05$).

their fatty acid composition, results in a variation of the total fatty acid composition. The best fit for our experimental results was obtained by a model in which grains with greater size contained a lower glycolipid proportion and higher relative amounts of neutral and phospholipids (Table 3). Although no experimental evidence is yet available, the suggested variations in lipid fraction proportions are very similar to those observed during the maturation of seeds. Thus, in developing wheat grains, decreases in relative amounts of phosphatidylcholine and diacyldigalactosylglycerol were accompanied by increases in the proportions of neutral and monoacylphosphatidylcholines [11]. Much of the latter lipid is located in the starchy granules where it represents *ca* 85% of the total lipid.

Two-rowed barley types have a higher TFA content compared to their six-rowed counterparts, due mainly to the higher absolute concentrations of 16:0, 18:1 and 18:2 (Table 4); winter barleys contain more 18:3 than spring barleys, which is in agreement with our previous findings [5]. Although direct genetic effects are involved in the observed differences between 2-rowed and 6-rowed barleys, it appears that the difference in grain size distribution pattern contributes to the explanation of some differences in fatty acid composition between the two barley types. Taking into account the greater weight proportion of bigger kernels in 2-rowed barleys than in their 6-rowed counterparts, the former are expected to contain more 16:0 and 18:1 and less 18:2 and 18:3. Thus, the higher 18:1 and the lower 18:3 proportions in 2-rowed barleys compared to 6-rowed barleys can be explained by the indirect effect of the genotype on fatty acid composition, namely the difference in grain size distribution between barley types and the relationship between grain size and fatty acid composition. The differences in 16:0 and 18:2 proportions can probably be ascribed to the direct effect of genotype on fatty acid composition.

EXPERIMENTAL

Mature seeds of nine varieties of barley were obtained from a field experiment by the Provinciaal Onderzoek-en Voorlichtingscentrum Rumbeke-Beitem (Belgium) during the 1984–85 growing season. The varieties represented six-rowed (Gerbel, W1261 and Corona) and two-rowed (Sonja, Marinka and Isabella) winter types and two-rowed spring types (Aramir, Triumph and Galant). All of these varieties were grown at the same location (Veurne, Belgium). Upon their arrival in our laboratory, the samples were stored in the dark at 5°. Each sample was fractionated according to grain width using a Steinecker separator (horizontal set of 3 sieves, 2.8, 2.5 and 2.2 mm, oscillating horizontally). The grains of width less than 2.2 mm were discarded.

Samples of seeds (± 50 g) were ground with an EBC-Casellamill to pass an 0.5 mm sieve. Moisture content of the flour was determined in triplicate by heating 5 g for two hr at 120°.

The content and composition of fatty acids, present as acylesters and unesterified fatty acids, were estimated in the flour by the micro-method of ref. [9], as described previously [5, 6]. The fatty acid Me esters (FAME) were analysed by FID GC using a stainless steel column (3 m × 3 mm i.d) packed with 10% SP2330 on chromosorb W AW 100/120 mesh. The column temp. was held isothermally for 4 min at 160°, followed by a programme from 160° to 190° at 1°/min. The N_2 flow rate was 22 ml/min, the injected sample size 1 µl. FAME were quantified

Table 3. Calculation of variations in total fatty acid composition, due to a modification of the neutral lipid (NL), glycolipid (GL) and phospholipid (PL) proportions (hypothetical model, see text)

			Fatty acid composition % of fraction				
Variety	Lipid fraction + proportion		16:0	18:0	18:1	18:2	18:3
Data							
Prilar [10]	NL	67.0	20.7	1.4	17.7	53.9	5.5
	GL	11.7	19.7	2.7	7.5	61.6	7.6
	PL	21.3	28.6	1.3	12.8	53.0	3.2
Bonanza [13]	NL	65.2	17.8	1.1	16.0	58.9	6.1
	GL	25.6	17.4	5.5	6.9	63.2	6.9
	PL	9.2	24.4	1.3	9.5	59.2	5.5

	Lipid fraction proportions			Estimated total fatty acid composition (%)				
Variety	NL	GL	PL	16:0	18:0	18:1	18:2	18:3
Calculated total fatty acid composition								
Prilar	70.0	20.0	10.0	21.3	1.6	15.1	55.4	5.7
	72.5	16.0	11.5	21.4	1.6	15.5	55.0	5.6
	75.0	12.0	13.0	21.6	1.5	15.8	54.7	5.5
	77.5	8.0	14.5	21.8	1.5	16.1	54.4	5.3
	80.0	4.0	16.0	21.9	1.4	16.5	54.1	5.2
Bonanza	70.0	20.0	10.0	18.4	2.0	13.5	59.8	6.2
	72.5	16.0	11.5	18.5	1.8	13.8	59.6	6.2
	75.0	12.0	13.0	18.6	1.7	14.1	59.5	6.1
	77.5	8.0	14.5	18.7	1.5	14.3	59.3	6.1
	80.0	4.0	16.0	18.8	1.3	14.6	59.1	6.0

Table 4. Mean grain size distribution (% wt), fatty acid proportions (% TFA) and fatty acid concentrations (mg/g dry wt) of two-rowed and six-rowed barleys

	Two-rowed	Six-rowed
Grain size distribution		
2.8 mm	77.2	50.6
2.5 mm	16.3	33.0
2.2 mm	4.4	12.0
Fatty acid composition		
16:0	23.6	25.1
18:0	0.8	0.9
18:1	12.2	11.4
18:2	54.2	51.0
18:3	6.6	6.9
Fatty acid concentration		
16:0	7.9	7.7
18:0	0.3	0.3
18:1	4.1	3.5
18:2	18.2	15.7
18:3	2.2	2.1
TFA	33.5	30.8

by the int. std. method. TFA is expressed as mg/g dry wt and fatty acid composition as % wt TFA. Analyses of variance were carried out using the SAS procedures ANOVA and GLM [12].

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